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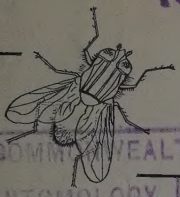

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Laboratory Studies on House Fly Populations

Raimon L. Beard

Introduction

The development of resistance to insecticides by many species of insects has dramatically focused our attention on the long-range effects of chemicals on insect populations. Resistance is certainly one possible effect of the continued use of insecticides. There must be many, more subtle, effects either favoring or opposing increase in insect numbers. The summary writings of Andrewartha and Birch (1954), Milne (1957), Nicholson (1955, 1958), Ripper (1956), Solomon (1957), Thompson (1956), and others (see Cold Spring Harbor Symposia vol. 22) emphasize the whole problem of population dynamics. In view of the controversial writings of these workers, we must consider as speculation the application of their conclusions to insecticides as factors in the insects' environment. It has become evident that more experimental work on populations subjected to insecticides is greatly needed. This bulletin introduces a series of studies directed towards this need.

A complete review of literature applying to this problem would be overwhelming. When results permit, it is planned to orient them with respect to findings of other workers.

Insect control by chemicals customarily follows two patterns. In one, the insecticide is applied as an emergency measure against an existing or developing population of excessive numbers of insects. In the other, the insecticide is applied to prevent an infestation of insects, regardless of the numbers then present or that might develop. If the insecticide is effective, the short-term results are easily recognized as a reduction in numbers of individuals or a protection from insect attack. The consequences of long continued use of insecticides with respect to fluctuations in insect numbers and the general biology of the target insect are difficult to assess. Nevertheless, many experimental ways are open.

In the work to be reported, the house fly, *Musca domestica* L. is the test insect. It is easy to rear in the laboratory, and it has a short life cycle. Its adaptability to changing environments is all too well known to those who would control its numbers. Not only does it continue to be a nuisance, but it remains a problem of economic and medical importance.

The experimental system is an attempt to copy, on a small scale, situations which can be found in natural populations. Several sites of infestation are presented. Food and breeding material are available at all times somewhere in the system, although they are not necessarily optimal at all times. The flies can reach directly or indirectly any site in the system, but the flies themselves, in accordance with their behavior in the particular environment, determine their own distribution. The experimental design purposely has elements of closely controlled laboratory situations and highly variable field conditions. It is believed to be more realistic than precise laboratory studies dealing with single factors, but more interpretable than field observations on population dynamics.

I. A Continuous Rearing System

The design of a laboratory experiment on the long-range effects of insecticides on house fly populations called for a continuous, self-regulating, rearing system. Easy maintenance and minimum disturbance were necessary. The system developed and described here has been in satisfactory use for over 20 generations. Its principal features may have application in other studies where flies without specified age or size are required.

Details contributing to easy maintenance include the use of disposable containers and easily measured food materials. Tight fitting parts of the rearing containers were designed to prevent fly or maggot escape—both to insure closed populations and to reduce the nuisance of escaped flies. The food media, modified from that suggested by Frings (1948), permits the assembly of the rearing units once each generation without further attention.

Twelve assembled rearing units are illustrated in Figure 1. Each unit consists of a container for larval food attached to a flight chamber containing adult food. The larval compartment consists of an eight-ounce paper cup lined with a polyethylene bag. This is stocked with

Figure 1. Assembly of 12 rearing units for studying closed populations of house flies.

50 grams of dried dog food (coarse meal) moistened with 50 ml. of a yeast suspension. This suspension is made up of one commercial package of active dry yeast (about 8 grams) in one liter of water. A paper cover placed in the cup over the medium prevents excessive drying and larval migration. A hole is punched in the cover through which flies can pass. The flight chamber is a plastic refrigerator box of about 40 cubic inches in volume. To the bottom of this is cemented a shallow circular plastic dish. The vertical edge of this dish serves as a rim or flange that just fits snugly into the paper cup. A hole drilled through both the bottom of the box and the attached dish allows flies to pass freely between the two sections.

In the flight chamber is placed a paper portion cup containing the adult food. This consists of dried milk powder and sugar, in equal amounts by volume. One tablespoonful of this mixture is provided. Water is supplied in an inverted bottle with a cotton dental roll wick. The box is ventilated by a screened hole in the top. A short length of rubber tubing, inserted in another hole in the box cover, connects other units in an assembly.

Flies oviposit in the larval food medium, and larvae develop as usual. Pupation occurs in the upper layers of the medium. The pupae and fly emergence are not disturbed.

The proportions and total quantity of food were found by trial to be satisfactory for production of sufficient numbers of flies; they are not necessarily optimal. Several hundred flies may develop in a single unit. If large numbers are present, the flies are small and obviously crowded.

In the system in use at present, four units are assembled as a battery. Each unit is connected above to a small plastic box which acts as a concourse for the four units (see Figure 2). Three batteries, intercommunicating through tubes via the concourse boxes, constitute a series (Figure 1). The three batteries in a series are managed in rotation. When flies are in peak emergence in the newest battery, the oldest battery is removed and a new one is set up at the opposite end. This means that one battery

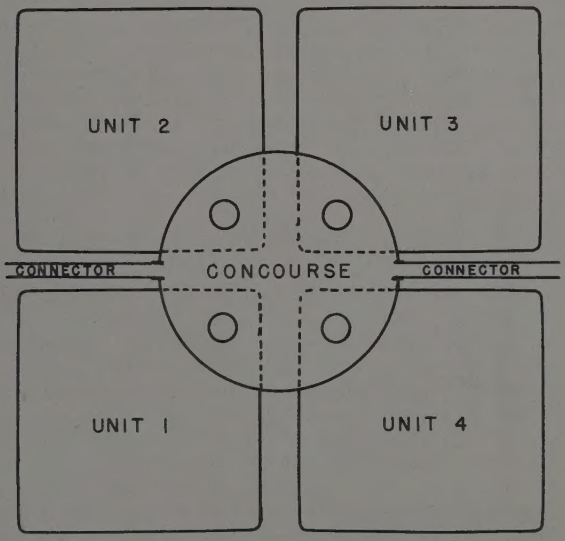

Figure 2. Assembly of four units and concourse box constituting a battery. Other batteries are joined by connecting tubes.

is left intact for almost three generations of flies. The food reserves in the discarded battery have been exhausted and are no longer attractive; the four units in this battery consequently harbor no developing flies.

A series of rearing units, therefore, is a microcosm confining a self-regulating population of flies. Each unit can be considered as a site

of infestation. The flies emerging in one unit can reach, by indirect routes, any other site in the system. Random activity and pressures of numbers assure dispersal. If one or more units are treated with insecticides, a local environmental hazard is introduced. Surviving flies are a mixture of those escaping treatment and those receiving sublethal doses. Because of the shifting population—both in numbers and space—counts of individuals would be rather meaningless. An indirect measurement of fly traffic is made instead. Papers of uniform size (45 sq. cm.) are fastened to one wall of each unit. At the time the units are taken down the papers are recorded as to degree of soiling. This is evaluated by homogenizing each paper in 100 ml. of water, filtering, and measuring light transmission of the filtrate with a colorimeter. Also fly size is estimated by weighing air dried samples of 50 flies from each battery.

II. Characteristics of Closed Populations Reared Continuously

The preceding section describes a rearing technique developed for laboratory study of the long-range effects of insecticides on house fly populations. Satisfactory application of the technique to this end requires that untreated populations develop according to a pattern from which deviations can be detected. This section reports on the trends shown in 20 generations of flies reared in seven sample populations, differing principally in the amounts of food available. No insecticides were used.

Methods

The basic rearing technique described in the preceding section was used with modifications to be described. The rearing units were originally stocked with flies obtained from the Entomology Section, Quartermaster Research and Development Laboratory, Natick, Mass. The initial number placed in each series was about 50 flies selected at random. Three generations were reared in order for the populations to build up and come to equilibrium within the limits of their environment. Data for the next twenty generations were then recorded. Extremes of temperature and humidity were reduced, but conditions were not kept constant. No special illumination was provided. The possibility of position effects associated with differences in light and temperature was reduced in two ways: (1) Twenty-four series of populations, including the seven reported here, occupied four tiers of six shelves. All series were rotated twice weekly. Thus in 12 weeks each series occupied every available position. (2) The four units constituting a battery, set up each new generation, were numbered consistently (Figure 2). In those series in which the four units were not alike, the different units were methodically shifted in relative position in each new battery.

The chief modifications of the basic arrangements were in reducing the quantity of larval or adult food or of eliminating larval food in certain units. When larval food was eliminated, moist sawdust was substituted. When larval food was reduced, 20 grams of dog food and 20 ml. of yeast suspension substituted for 50 of each. When adult food was reduced, one teaspoonful substituted for one tablespoonful. A further modification, in two series, was the substitution of a portion of the flies in the 16th generation by flies obtained from Dr. H. H. Moorefield, of the Boyce Thompson Institute. The purpose of this was to introduce different genetic elements into the breeding stock. The constitution of the seven units is indicated in Table 1.

An analysis of variance was made of the traffic indexes from each series to differentiate the variation between the four units in each battery, and between the twenty batteries. Also, fly size was analyzed to detect significant differences among the seven series and batteries of each series.

Results and discussion

In the three-battery rearing system, the flies at any time can represent one, two, or three generations. Because of this and because of differences in developmental time among the flies, a given population

Table 1. Food complement in the four units of each battery in seven experimental populations (100 = full amount of appropriate food)

Series	Number of units	Larval food	Adult food
A	4	100	100
B	3	100	100
	1	0	100
C	2	100	100
	2	0	100
D*	4	100	100
E*	2	100	100
	2	0	100
F	4	40	33
G	2	40	33
	2	0	33

* In D and E, flies from a different source were added in the 16th generation.

level could be constant. This has not been evident, and distinctive generations are indicated by a rise and fall in numbers associated with a life cycle of about 2 to 2½ weeks. This is undoubtedly fostered by the technique, as new sites of infestation (new batteries) are introduced at peak emergence of a generation. Even so, some flies are present at all times, although they are of different ages and of overlapping generations.

As previously mentioned, estimates of actual numbers of flies would be impractical. Nevertheless, the index of fly population, based upon the extent of soiling as described, evaluates very well the fly traffic over the area provided. It includes, however, contributions of three generations of flies: the flies invading a new site, the flies developing in and emerging from this site, and flies of the next generation wandering back into this older unit. Of these, the developing flies contribute most to the traffic estimate. It has not been feasible to establish a calibration curve for equating a traffic index with known numbers of flies for a definite length of time. The low end of such a curve can easily be obtained, but the high end cannot; known numbers of flies, of more than a hundred or so, are difficult to maintain for extended periods. As a point of reference, however, the amount of soiling from 100 flies in 10 days has given a traffic index reading of 19. Numerous readings for single units have exceeded 1000. Although this represents traffic for six weeks or more, several hundreds or even thousands of flies are obviously contributing to papers registering such high values.

A supplemental reading for fly traffic was obtained by washing the concourse box with 250 ml. of water, filtering, and measuring the optical density of the filtrate. In general, these readings were low and much more uniform among the different series than were the unit readings. The concourse box readings, therefore, are not particularly discriminating.

The population trends of the seven series are graphically illustrated in Figure 3. Each bar represents the sum of the fly traffic indexes for

the four units and the concourse box. In Figure 3, each bar is labelled as a generation. This is not strictly correct, for, as has been mentioned, flies of three generations contribute to the index. To what extent this

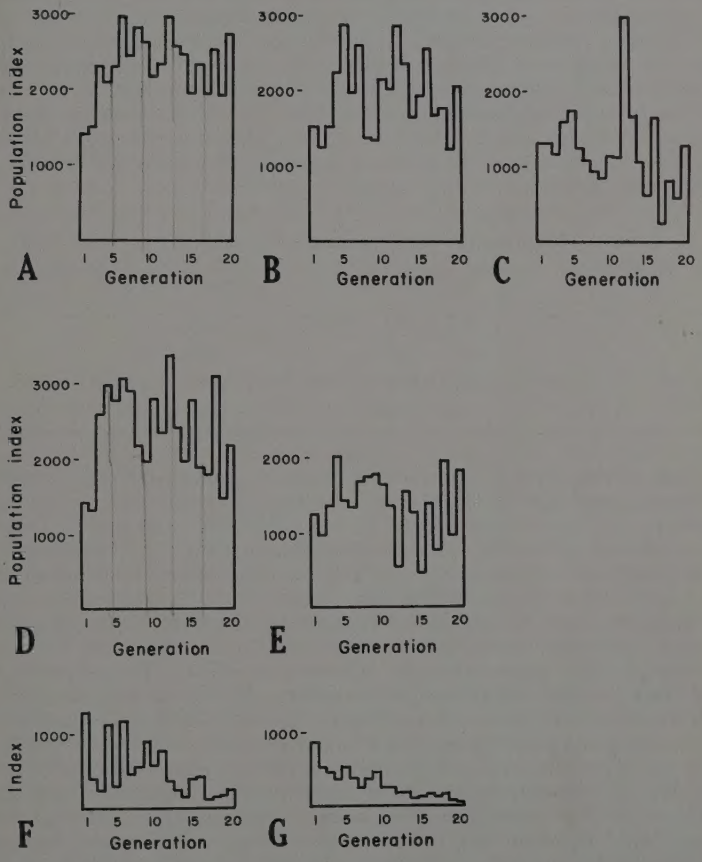

Figure 3. Population trends of flies in seven series maintained 20 generations; judged by measurement of fly traffic.

equalizes the readings cannot be stated, but further mention of this will be made.

The series D and E show no marked effect of the introduced flies. Apparently detrimental inbreeding, if any, had not occurred to the point where additional genetic material served as a stimulus to greater reproduction. This means that Series A and D are essentially alike, as are C and E. The analyzed data are consistent with this conclusion.

In series A and D the data were tabulated so that any differences among the units would indicate the effect of the position occupied by

the units. Thus the mean traffic indexes for units 1, 2, 3, and 4 were respectively 673, 567, 540, and 582 in series A, and 555, 559, 570, and 633 in series D. These values do not differ significantly. From this it can be concluded that although rotation of treated units may be desirable as a precaution against position effects, it appears to be unnecessary.

The data from series B, C, E, and G give an estimate of fly dispersal from breeding areas. No flies developed in those units where moist sawdust substituted for larval food, so any fly traffic in such units represented flies migrating from breeding units. Table 2 shows that whereas dispersal did occur, it was not sufficient to equalize the population in all units. In other words, the traffic index is strongly biased by the numbers of flies developing in a unit.

Table 2. Mean traffic index in units with and without larval food

	With larval food			Without larval food	
B	675	565	591	117	
C	495	507		100	121
E	598	608		99	111
G	69	106		39	28

It is obvious from Figure 3 that the several series differed materially in their generation to generation variation. The relative uniformity in series A, D, and to lesser extent B, was somewhat unexpected. It would be predicted from the work of Nicholson (1955) that self-regulating populations like these would be cyclic, with rather wide oscillations. The build-up of heavy populations should lead to competition among both larvae and adults. This should cause a decline in the population until competition would be relaxed, thereby permitting the population to increase. The expected cycles did not materialize. The population indexes show more or less uniform numbers at high levels. As has been stated, the index would be leveled to some extent by the contribution of overlapping generations. This, however, does not fully explain the absence of cyclic trends. It was shown that dispersal does not equalize the distribution of flies among the different units when the flight chambers are in the same condition, so it is unlikely that dispersal would equalize fly numbers between old and new units. Moreover, observation confirms that in every generation reported here, the unmodified series were crowded with flies. In other words, the observed fluctuations represented differences in degrees of crowding rather than differences between low and high populations. In series F and G food was expected to, and indeed did, limit the population. There is every reason to believe that in the other series, food also was limiting, but at higher population levels. Because of this limitation, competition did occur, and it was sometimes observed that when larval food became depleted, the later developing larvae failed to complete their growth. The competition was not so devastating as to cause a subsequent drop in population to low levels.

Series C, E, and F show fluctuations from rather low to high population levels, and, at times, cycles are suggested. These are not consistent nor of the sort predicted by observations of Nicholson (1955) on blow-

fly populations. All of the other series show greater variation from the mean populations than series A and D. From these and other data not here reported, it appears that a modified portion of the total environment causes greater fluctuations from generation to generation than when all breeding sites are uniform.

Series G shows a progressive reduction in fly numbers which, if continued, would lead to extinction. A possible cause of this could be an increasing inbreeding and a consequent loss of fertility. This was disproved by restoring the full complement of food for both adults and maggots. Figure 4 shows that when provided with adequate food, the marginal population, present in the twentieth generation, built up in two or three generations to fly numbers consistent with the standard series in which adequate food had been present during the entire period. Fly numbers in series F likewise decreased progressively, although the

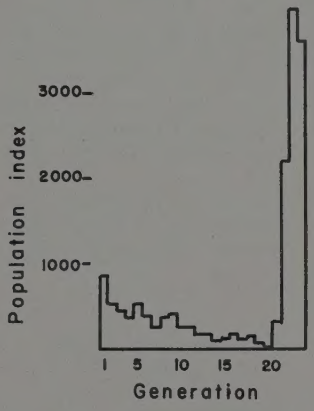

Figure 4. Population trend of flies in Series G. Adult food and rearing medium were restricted for 20 generations. For four subsequent generations more adequate food and rearing medium were provided.

trend was less regular than in series G. These flies, too, promptly developed in large numbers when the full complement of food was provided after the twentieth generation. Not attributable to loss of fertility, the decline in fly populations probably results from inadequate dispersal of flies to breeding sites when the food medium was at its best. Small amounts of larval food dried much faster than the standard amount, and if oviposition was delayed until the food medium became unsuitable, few or no flies developed. In series G, where half of the sites, those with sawdust, were unsuitable for breeding, random dispersal further reduced the chance of fertile flies reaching favorable breeding media.

Fly size (weight) did not differ significantly, either among the seven series or from generation to generation. This conclusion does not apply to data at hand for other series in which fly size is more variable. It is, therefore, concluded that if fly size is a measure of competition, competition was rather uniform in the seven series here reported.

Summary

As an introduction to a series of laboratory studies on the long-range effects of insecticides on house fly populations, this bulletin describes rearing techniques and the characteristics of untreated populations maintained for 20 generations.

The rearing of a closed population of flies is continuous. New breeding sites are added and exhausted breeding sites are removed in rotation. Twelve intercommunicating rearing units constitute a closed system. Flies can reach directly or indirectly any site in the system and are free to choose where they go. Food and breeding material are provided only once each generation. Easily measured and easily prepared food, and the use of disposable containers, facilitate maintenance. Fly traffic, rather than fly numbers, is used as an index of population size.

With adequate food and uniform breeding sites, large numbers of flies develop every generation. Lack of uniformity in breeding areas seems to aggravate fluctuations in fly numbers from generation to generation. Except for differences in fly numbers associated with the life cycle, no consistent cyclic fluctuations have been evident.

In two series in which food and breeding media were severely limiting, populations steadily declined. Extinction was probable, but adequate food and breeding media permitted prompt recovery of populations to high levels.

As judged by the lack of effects of introducing outside flies, no deleterious inbreeding had occurred in 16 generations.

In the seven control series reported, fly size was uniform between series and between generations.

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